# PROBIOTIC (LBC) IN BUFFALO HEIFERS RATION: 2- EFFECT ON SOME BLOOD PARAMETERS

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#### SUMMARY

To study the effect of lactobacillus concentrate (LBC) on some blood parameters, a total of 23 buffalo heifers divided into 4 groups (G1,G2,G3 and G4) were fed a basal diet supplemented with 0, 6.5, 13 or 26 g LBC/day. The basal diet consisted of concentrate feed mixture and corn (70%) plus berseem hay and rice straw (30%) as roughage. Blood sampling were collected once monthly starting from six up to 18 month of age.

The results showed that animals supplemented with the level of 13 g LBC/day had the highest (P<0.05) level of plasma protein. In the same time, differences among groups in plasma albumin content were not significant, while differences in plasma globulin among different groups were significant (P<0.05) and were highest for the animals fed ration supplemented with 13 g LBC. Both GPT and GOT activity were not affected significantly with LBC supplementation.

Keywords: Lactobacillus, buffalo heifers, blood and parameters

### INTRODUCTION

In Egypt, the buffalo is considered the main dairy animal in addition to its significant contribution to the annual beef production (42-45%). During the past 20 years, various feed additives and subcutaneous implants have been used to stimulate growth and improve feed

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utilization of different ruminant species except the buffalo (Bechman et al., 1977; Berger et al., 1981, Spires et al., 1990; Brandt et al., 1991 and Zinn and Borques, 1993).

Lactic acid bacteria (Streptococcus bovis and a rumen Lacto-bacillus sp.) are the first bacteria to be found in the weaned lamb (Hobson, 1976). Viable culture have been used to reduce the incidence of diarrhoea in dairy calves (Bechman et al., 1977).

Commercial production of lactobacillus culture concentrate has now entered a more a reputable era. It has been used to prevent scouring in calves (El-Garhy, 1982) by producing acidic media in the rumen which is unsuitable for the normal growth of the bacteria which cause diarrhoea.

This trial was conducted to study the effects of lactobacillus concentrate (LBC) on some blood parameter of buffalo heifers.

## MATERIAL AND METHODS

The trial was carried out on buffalo heifers from the herd of milk replacer research center, Faculty of Agriculture, Ain-Shams University. The field experiment extended for 14 months, e.i, till animals reach 18 months of age.

A total number of 23 female buffalo calves about four months old and weighing 92-95 kg (at the beginning of the experiment) were used. The animals were divided into four groups. The groups were randomly assigned to the following treatments:-

- Group (1) (n=6): Animals received their ration without supplement of LBC (control).
- Group (2) (n=6): Animals received the control ration in addition to 6.5 g/ head/ day LBC supplement.
- Group (3) (n=5): Animals fed the control diet in addition to 13 g/head /day dose of LBC.
- Group (4) (n=6): Animals received the control ration in addition to 26 g/head/day dose of LBC.

Feed ingredients and allowance were as described in the first paper of this series (El-Ashry et al., 1993).

Blood samples (10 ml) were obtained by using dry evacuated test tubes and double needle from the external.

jugular vein of 15 animals, three animals from each of groups 1, 2 and 3 and six animals from group 4. The blood samples were transferred in ice box until centrifuged for 15 minutes at 3,000 r.p.m. The obtained plasma were transferred into 5-7 ml vials and stored at -20°C till analyzed. Blood sample were collected once monthly in the morning before concentrate feeding and watering.

Plasma total protein was determined by a calorimetric method using the biuret reagent as described by Armstrong and Carr (1964). The determination of plasma albumin was carried out according to the method of Doumas et al. (1971). The concentration of globulins for each plasma sample was calculated by subtracting the albumin concentration from the total protein concentration. The determination of cholesterol was carried out according to the method of Watson (1960). Plasma transaminases (GOT and GPT) were determined calorimetrically by the method of Reitman and Frankel (1957).

The least square procedure (Harvey, 1960) was followed for statistical analysis.

# RESULTS AND DISCUSSION

# Plasma Protein

Total protein (TP) in blood plasma of growing buffalo heifers at different ages are shown in Table 1. Total proteins was high (P<0.05) for G2 (6.5 g LBC/day/head) during different intervals, followed by G1, (control group). The lowest TP values were recorded for the fourth group. For the entire experimental period, G2 showed the highest value (6% over the control) while G4 had a value representing 96% of the control group. Statistical evaluation for the values obtained during different experimental intervals showed significant differences during the third period (P<0.01) and slightly significant in the fifth interval (P<0.05). In the other intervals, the differences were not statistically significant (P> 0.05).

Lowest total protein content of blood plasma was detected during the first interval (at the  $6\underline{th}$  month old). It then increased gradually to reach the maximum at the fourth interval (P<0.01) i.e when the animals were at 15 month old.

Table 1. Mean of total blood plasma proteins (TP), albumin (A), globulin (G), and albumin/ globulin ratio (A/G) of buffalo heifers fed a ration supplemented with different levels of LBC

Groups		Heifers age ( months ) 0						
		6	9	12	15	18	group	
	TP	8.0 ±.29	7.7 ±.05	8.1 <sup>A</sup> ±.13	8.6 ±.15	8.0°±.11	8.1	
G1	A	4.6 ±.10	4.5 ±.04	5.4 ±.15	$5.3 \pm .13$	5.1 ±.18	5.0	
	G	3.5 <sup>A</sup> ±.19	3.2" ±.08	2.7 <sup>A</sup> ±.08	3.3 <sup>^</sup> ±.13	2.9°±.14	3.1	
	A/G	1.3 <sup>^</sup> ±.03	$1.4^{a} \pm .05$	1.9 <sup>A</sup> ±.09	1.6 <sup>A</sup> ±.08	1.8 ±.14	1.6	
	TP	8.1 ±.19	8.7 ±.47	8.9 <sup>B</sup> ±.24	8.8 ±.35	8.5 <sup>b</sup> ±.11	8.6	
G2	A	4.5 +.29	4.6 ±.21	5.0 ±.14	5.1 ±.02	4.9 ±.16	4.8	
	G	3.6 <sup>A</sup> ±.19	4.1 <sup>h</sup> ±.33	3.9 <sup>B</sup> ±.14	$3.7^{8} \pm .33$	3.6 <sup>h</sup> ±.08	3.8	
	A/G	1.3 <sup>A</sup> ±.13	$1.1^{b} \pm .08$	1.3 <sup>B</sup> ±.04	1.4 <sup>^</sup> ±.11	1.4 ±.07	1.3	
	TP	7.6 ±.30	8.2 ±.05	7.4°±.04	8.3 ±.12	7.7°±.16	7.8	
G3	A	4.5 ±.13	4.6 ±.22	5.0 ±.22	5.6 ±.06	5.0 ±.23	4.5	
	G	$3.1^{8} \pm .38$	3.6° ±.18	2.4 <sup>4</sup> ±.23	2.7°±.07	2.7°±.26	2.9	
	A/G	1.5 <sup>A</sup> ±.09	$1.3^{ab} \pm .13$	2.1 ±.22	$2.1^{B} \pm .04$	1.9 ±.24	1.7	
	TP	7.2 ±.22	7.9 ±.32	7.8 <sup>A</sup> ±.17	8.2 ±.14	7.8°±.18	7.8	
G4	Α	4.6 ±.18	4.7 ±.16	5.2 ±.16	5.4 ±.18	5.1 ±.20	5.0	
	G	$2.6^{\circ} \pm .07$	3.2° ±.18	2.6 <sup>A</sup> ±.21	2.7 <sup>C</sup> ±.16	2.7°±.22	2.8	
	A/G	$1.7^{B} \pm .06$	1.5 <sup>ac</sup> ±.06	2.1 ±.21	2.0 <sup>8</sup> ±.15	2.0 ±.23	1.9	
	TP	7.7	8.1	8.1	8.4	8.0		
	A	4.5	4.6	5.2	5.3	4.9		
	A** G~	3.2	3.5	2.9	3.1	3.0		
	A/G <sup>**</sup>	1.5	1.3	1.9	1.8	1.8		

<sup>\*</sup> mg/100 ml plasma.

Mean values of plasma albumin (Table 1) were similar for different heifer-groups during the experimental period. However, these values increased steadily and gradually till they reached maximum level after 12 months of the start of experiment (18 month old), being 118% of the first interval.

Changes in plasma albumin concentration between each two successive experimental intervals had no consistent trend. A marked jump was recorded between the second and the third growth interval (12%); percentage differences

<sup>\*\*</sup> Overall mean.

a,b,c & A,B,C values different superscripts in the same column for the same
parameter are significantly different (P<0.05 and P<0.01)
respectively.</pre>

between the fourth and the third interval was only 3% followed by a negative value between the fifth and the fourth experimental intervals (namely - 8%).

With regard to total plasma globulin content (Table 2), it was noted that overall mean value of 6.5 g LBC treated heifers, G2, was higher by 21% than the control group. However the corresponding values of G3, and G4, were 93.2 and 88.7% of the value of the control group, respectively, the differences were statistically significant (P<0.01).

Table 2. Mean of blood plasma cholesterol\* (CH) and transaminases\* (GPT and GOT) of buffalo heifers fed ration supplemented with different levels of LBC.

Gro	ups	Heifers age ( months )							
01 0	rups	6	9	12	15	18			
Histo	СН	82.5°± 6.4	75.4±15.1	91.5 ± 9.3	97.6 ± 1.7	91.4 ± 2.3			
G1	GPT	21.8 ± 4.9	23.1± 2.6	27.4°± 5.3	31.3 ± 5.5	28.3 ± 4.0			
	GOT	134.1 <sup>A</sup> ± 7.0	143.1±22.1	152.2 ± 6.5	102.7 <sup>A</sup> ± 6.7	117.5°± 7.2			
	СН	68.3 <sup>h</sup> ± 2.1	65.1± 3.5	89.7 ± 5.7	87.2 ± 5.0	93.8 ± 5.0			
G2	GPT	29.0 ± 5.2	18.8± 5.2	46.6°± 4.7	34.8 ± 2.2	38.9 ± 2.9			
	GOT	183.2 <sup>B</sup> ±10.4	146.3±14.9	172.9 ±12.3	182.5 <sup>8</sup> ± 9.5	172.2 <sup>h</sup> ±10.2			
	СН	69.8 <sup>h</sup> ± 4.0	72.2± 9.2	85.7 ± 5.5	86.8 ± 7.6	90.1 ± 3.8			
G3	GPT	22.8 ± 2.7	26.2± 3.5	35.7°± 1.0	36.0 ± 4.0	39.0 ± 3.0			
	GOT	182.2 <sup>H</sup> ±11.3	154.0±26.0	124.0 ±22.6	119.3 <sup>A</sup> ±18.8	133.7°±17.6			
	СН	64.7 <sup>h</sup> ± 1.9	82.5± 5.5	86.1 ± 4.3	91.5 ± 4.2	85.3 ± 3.4			
G4	GPT	23.7 ± 3.9	19.4± 3.1	29.2"± 3.0	31.7 ± 2.5	33.0 ± 3.0			
	GOT	170.0°± 3.9	170.0±11.0	169.0 ±7.55	157.6°±10.3	157.3 <sup>h</sup> ± 9.9			
	CH.	71.33	73.81	88.40	90.86	77.62			
	GPT ***	24.33	21.88	34.73	33.45	34.80			
	GOT	16	15	15	140	145			

<sup>\*</sup> mg/100 ml.

This lack of differential effect of LBC level may indicate that, the level of LBC affect the total plasma globulin content. However, appreciable changes were noted and was related to time of sampling.

<sup>\*\*</sup> unit /100 ml.

<sup>\*\*\*</sup> Overall mean.

a,b,c & A,B,C values different superscripts in the same column for the same parameter are significantly different (P<0.05 and P<0.01), resp.

Values for albumin: globulin (A/G) ratio (Table 2) between groups were insignificant.

The present finding of plasma protein mean values were between 7.8-8.6 g per 100 ml (Table 2). However, mean values of albumin were 4.8 to 5.0, while globulin mean values were between 2.8-3.8, g/100 ml respectively. Values obtained in the present study were higher than those reported by Hussein (1986) on male buffalo calves for serum total protein  $(5.6-7.7~\rm g\%)$ ,  $(2.9-3.55~\rm g\%)$  for serum albumin and  $(2.5-4.36~\rm g\%)$  for serum globulin. This could be attributed to the fact that the animals of this study consumed more CP than those of Hussein (1986). Also, our values were in line with ranges reported by Rowlands et al. (1974) for total protein, albumin and globulin in cattle which were: 4.34-8.49, 1.88-3.25 and 2.46-5.24 g/100 ml plasma.

The high globulin concentration recorded in the present study are in agreement with those reported by Hussein (1986), who attributed this increase mainly to the increase in globulin and development of the immune system.

The data also revealed highly significant effect of age on plasma total protein, albumin and globulin in all groups. This finding is in agreement with that reported by Gartner et al. (1966), Hegns (1971); Kitchenham and Rowlands (1976); Green et al., with cattle (1982) and Hussein (1986) working with growing buffalo.

# Plasma Cholesterol

Data of Table 2 show that addition of different levels of LBC to buffalo heifers rations had no effect (P>0.05) on blood serum cholesterol of the experimental animals throughout 14 month experimental period.

Generally, values of serum cholesterol obtained in this study were higher than those of El-Sayed (1991) who reported values ranged 52-64 mg/100 ml serum for male buffalo calves at the same ages. Moreover, our values were lower than those of buffaloes recorded during the stages of oestrous cycle (142-303 mg/100 ml) which were reported by Chaudhry and Ahmed (1973).

On the other hand, by age progress, blood serum cholesterol of all the experimental animals increased significantly (P<0.01). The same observation was noticed by Tumbleson and Hutcheson (1971) and O'Kelly (1973).

#### Plasma Transaminase

The activity of plasma glutamate-pyruvate transaminase (GPT) was slightly increased in all groups with the progress of body growth (Table 2). The use of LBC significantly (P<0.05) increased GPT activity.

On the other hand, no consistent pattern was noticed in the plasma glutamate oxaloacetate transaminase (GOT) activity with advanced age in any of the four groups. However, as a function of treatment in the first growth interval, the treated groups with LBC had higher values of GOT than the control group (P<0.01). Within treatments, both 6.5 g LBC and 13 g LBC/head/day were likely similar and higher than those of 26 g LBC/head/day (P>0.01).

Activities of both GPT and GOT were significantly affected by age (P<0.01) and also the interaction between age x treatment. Thus age and age by level of LBC caused changes in the activity of plasma GPT and GOT.

The present finding showed higher ranges for GOT (129-171 unit %) than those reported by Hussein (1986) with male buffalo calves (72-115 unit %), and by Zafer et al. (1979) using Egyptian native cattle of 2.5 years old (23.7-76 units/ml). However, GOT activity reached a peak at 9-12 months of age and was slightly decreased with the progress of age thereafter. Such pattern is in agreement with that reported by Hussein (1986).

On the other hand, the observed range for GPT (26.4-33.6% units) confirmed the data of Hussein (1986) who reported 34-58 units%. However, the observed increase in GPT activity was not in agreement with what was found by Hussein (1986), who noticed that GPT activity reached a peak value at the 3rd month on experiment then decreased gradually till the end of the experiment. Differences in estimates might be due to differences of sex or age.

Results obtained by the same authors conducted with add LBC to buffalo heifers rations showed that addition of 6.5 g LBC/day increased feed intakes and daily gain which was reflected on age at conception (see part 1, El-Ashry et al., 1993).

It was concluded that a level of 6.5 g/head/day LBC during the first 6 months gave best result with regard to protein metabolism at the blood level.

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منشطات النمو LBC في علائق عجلات الجاموس: ٧- التأثير على مكونات الدم

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لدراسة تأثير منشط النمو (اللاكتوباسياس) المركز LBC على بعض مكونات الدم في عجلات الجاموس استخدم ٢٣ عجلة (عمر ٤ شهور) جاموس قسمت الى أربعة مجاميع تغذى المجموعه الاولى - مجموعة المقارنة - على عليقة قاعدية تحتوى على مواد مركزه (علف موحد وذره صفراء) ومواد خشنه (دريس برسيم وقش ارز) في حين تغذت المجموعة الثانية والثالثة والرابعة على العليقة القاعدية بالاضافة الى ٦,٥ و ١٣ و ٢٣ جم مركز LBC يوميا لكل رأس.

ثم أخذ عينات دم من كل حيوان ابتداء من عمر الشهور وحتى عمر ١٨ شهرا مرة واحدة شهريا بعد الوجبه الصباحية لدراسة تأثير المعاملة على مكونات الدم من البرونين الكلى والالبيومين والجلوبيولين والكلسترول.

وأظهرت النتائج أن مستوى بروتين الدم ارتفع معنويا للمجموعة المغذاه على ١٣ جم LBC يوميا في حين أن الاختلافات في البيومين السيرم للمجاميع المختلفه كانت غير معنويه أيضا أظهرت النتائج ارتفاعا معنويا لمستوى جلوبيولين سيرم دم الحيوانات المغذاه على مستوى ١٣ جم LBC يوميا . لم تتأثر انزيمات CPT, GOT معنويا بالمعاملة بمنشط النمو LBC .